

HPLC Application

ID No.: 18893

Albumin and IgG (2:3) on BioSep2000 (2)

Column: BioSep[™] 5 μ m SEC-s2000 145 Å, LC Column 300 x 7.8 mm, Ea

Dimensions: 300 x 7.8 mm ID

Order No: 00H-2145-K0

Elution Type: Isocratic

Eluent A: 100mM Phosphate buffer \pm 200mM Arginine pH 6.8

Gradient Profile:	Step No.	Time (min)	Pct A
	1	0	100

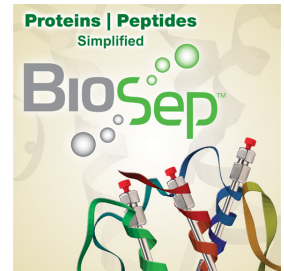
Flow Rate: 1 mL/min

Col. Temp.: ambient

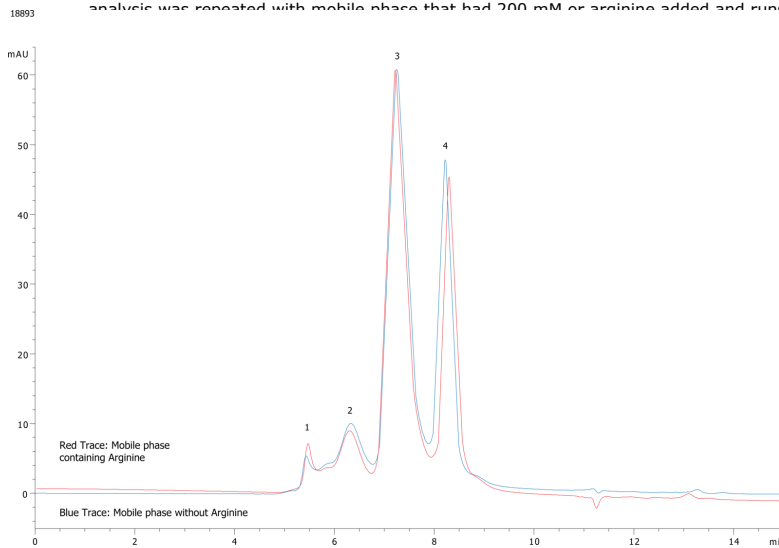
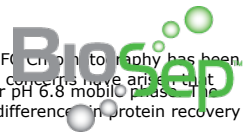
Detection: UV-Vis Abs.-Variable Wave.(UV) @ 280 nm (ambient)

Analyst Note: Application Topic: Inertness of GFC phases and accurate aggregate analysis

Protein aggregation is the post translational modification of most interest for those developing protein therapeutics. GFC (and HPLC) has been the "gold standard" method for quantitating aggregates in therapeutic proteins for over twenty years, however recent concerns have arisen about it. In this application a mixture of Ig-G and albumin was analyzed using a BioSep 2000 using a 100 mM phosphate buffer pH 6.8 mobile phase. The analysis was repeated with mobile phase that had 200 mM of arginine added and runs were overlaid. Any significant difference in protein recovery



Products used in this application:



ANALYTES:

- 1 aggregate
- 2 IgG dimer
- 3 IgG monomer
- 4 BSA monomer peak